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Review

Inhalation solutions — Which ones may be mixed? Physico-chemical compatibility of drug solutions in nebulizers — Update 2013

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Abstract

Many patients suffering from chronic respiratory diseases rely on inhalation therapy with nebulizers. About 25% of patients who need to inhale several different drugs per day save time by mixing them for simultaneous inhalation. This review presents a comprehensive overview of the available data concerning physico-chemical compatibility of commonly mixed nebulizer solutions and suspensions. Information is based on our in vitro studies and a thorough literature search.

Results indicate that many nebulizer solutions/suspensions are mixable without provoking incompatibilities. However, certain excipients contained in some of the tested drug products could be identified as a reason for incompatibilities, e.g. impaired activity of dornase alfa. Studies assessing the aerosol characteristics of compatible mixtures nebulized with commonly used nebulizers are limited and should be encouraged. The clinical efficacy of simultaneous inhalation of duplicate, tripartite or quadripartite mixtures must be evaluated in clinical studies before final recommendations for the inhalation regimens can be made.

Keywords: Nebulizer solution/suspension; Compatibility; Mixture; Aerosol; Review

Contents

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1. Introduction

Patients suffering from cystic fibrosis (CF) or other chronic airway diseases widely rely on inhalation therapy. Besides dry powder inhalers (DPI) and metered dose inhalers (MDI) nebulizers are used, although nebulization is much more time consuming than inhaling with a DPI or MDI. In contrast to DPI's and MDI's, nebulizers deliver the drugs continuously and larger doses can be administered. For children nebulization is advantageous because the coordination requirements are less strict [1]. They often cannot handle a MDI since the coordination of medication release (i.e. actuation of the device) and simultaneous inhalation is too difficult. Concerning DPI's, children aren't able to generate the peak inspiratory flow (PIF) necessary to release the drug [2].

Studies indicate that different nebulizer systems from different manufacturers, i.e. ultrasonic, jet, or vibrating membrane nebulizers with ventilator, humidifier or Y-piece, generate different aerosol mass distribution profiles and thus, drug deposition in the airways varies [3–8]. Adequate drug delivery can be achieved if a specific medicinal product approved for nebulization is prescribed together with a specific nebulizer which was evaluated to be an effectively functioning drug-device combination [5]. But apart from that, daily practice is even worse, especially if patients have to inhale several drugs per dose interval. Instead of cleaning, reassembling and refilling the nebulizer to perform consecutive nebulization at least 25% of patients save time by mixing the inhalation solutions/suspensions [9,10]. Nebulization of medication mixtures increases the aerosol output of the nebulizer while its dead volume remains the same. If nebulization is continued until the nebulizer runs dry, total mass output and inhaled mass of the nebulized drug will rise [11–14]. However, incompatibility and/or instability of the medication mixtures can lead to impaired drug safety and/or reduced efficacy up to treatment failure.

Further, simultaneous nebulization of inhalation solutions can affect drug delivery by altering the aerosol particle size distribution. Particle size should be 1 to 5 μm in diameter, because larger particles will deposit in the upper airways and smaller particles may be exhaled [15]. Hence, even if physico-chemical compatibility of mixtures is proven final recommendations for simultaneous inhalation cannot be made. Aerodynamic characteristics of mixtures need to be studied. In addition, the clinical relevance of inhaling different drugs simultaneously and the differences in therapeutic outcome compared to consecutive inhalation should be investigated.

Drug substances commonly prescribed for nebulization therapy in Europe comprise the bronchodilators albuterol (salbutamol) and ipratropium bromide, the corticosteroids budesonide and fluticasone-17-propionate, the antibiotics tobramycin and colistimethate, and dornase alfa. In addition, formoterol, acetylcysteine, sodium chloride solutions (0.9% to 7%), and the mast cell stabilizer cromolyn are used.

In 2006 we published our first results concerning physico-chemical compatibility and stability of several nebulizable drug solutions/suspensions predominantly used in Europe [10]. Since then various mixtures have been experimentally tested by us and other groups, also in regard to the resulting aerosol characteristics. Results were published in several articles of the primary literature and Burchett et al. developed a preliminary compatibility guide for aerosolized medications used in the US [16]. In this issue we present a comprehensive overview of the available data based on our own studies and the search of relevant literature as well as the up-to-date version of the compatibility data in table format.

2. Methods

2.1. Physico-chemical compatibility of nebulizable drug mixtures

In the following paragraphs we present an overview of the methods used to determine the physico-chemical compatibility of duplicate, tripartite and quadripartite drug solutions/suspensions inhaled via nebulizers. According to the conventional definition of compatibility, mixtures of inhalation medications can be designated as physico-chemical compatible, when chemical stability (≤ 10% degradation) of each active substance and unchanged pH values, osmolality and physical appearance are given over a test period of ≤ 24 h. In general, potencies or concentrations of the active substances of the mixtures were determined under specified conditions.

2.1.1. Preparation of admixtures

Test solutions/suspensions were prepared by mixing commonly used doses of medication approved and marketed in

Europe. Mixtures were stored at room temperature or under refrigeration and exposed to light for several hours. Analyses were performed immediately after preparation and after defined intervals [17–20].

2.1.2. Physical compatibility

Physical compatibility of the inhalation mixtures was determined by visual inspection for any changes up to 24 h after mixing as well as by measuring pH and osmolality [17–21]. Mixtures were designated as physically incompatible when color or odor changed, and haze or precipitation occurred. The lack of physical incompatibility does not rule out chemical decomposition.

2.1.3. Chemical compatibility (Table 1)

Potencies of antibiotics in inhalation mixtures were determined by fluorescence immunoassay (tobramycin) [18,21] or by using the 'Microbiological assay of antibiotics' (agar diffusion assay) of the European Pharmacopoeia (Ph. Eur.) in comparison to a reference standard (colistimethate, tobramycin) [17,22].

Dornase alfa activity was determined by a kinetic colorimetric DNase activity assay [18,20]. Stability of dornase alfa was determined by size-exclusion high performance liquid chromatography (SE-HPLC), ultraviolet spectroscopy, sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) and tentacle strong cation-exchange chromatography [21].

Concentrations of the corticosteroids (budesonide and fluticasone-17-propionate) and the bronchodilators (ipratropium bromide and albuterol) were measured by stability-indicating high performance liquid chromatography assays with ultraviolet detection (RP-HPLC) [17,19,20,22,23].

2.2. Aerosol characteristics

A subset of mixtures was analyzed in regard to their aerosol characteristics after nebulization with the PARI eFlow® rapid. Particle size distribution, Mass Median Aerodynamic Diameter (MMAD), Geometric Standard Deviation (GSD) and Fine Particle Fraction (FPF; particles <5 μm) were determined via cascade impaction with the Next Generation Pharmaceutical Impactor (NGI) in comparison to aerodynamic parameters of unmixed solutions under specified conditions (Fig. 1). RP-HPLC and SE-HPLC were used to assess drug concentrations deposited on each impactor stage, in the induction port and in the nebulizer. We tested duplicate mixtures of dornase alfa and tobramycin, i.e. Pulmozyme® with Bramitob® and Pulmozyme® with TOBI® (Table 3) as well as the tripartite mixture of ipratropium bromide, albuterol and fluticasone-17-propionate, i.e. Atrovent®, Sultanol® forte and Flutide® forte.

Table 1
Analytical methods (own data only) used to determine chemical compatibility of mixed nebulizer solutions/suspensions.

Drug	Method
Dornase alfa	Kinetic colorimetric DNase activity assay Size-exclusion high performance liquid chromatography (SE-HPLC) Ultraviolet spectroscopy Sodium dodecyl sulfate polyacrylamide gel electrophoresis (SDS-PAGE) Tentacle strong cation-exchange chromatography
Tobramycin	Fluorescence immunoassay, microbiological assay of antibiotics Ph. Eur.
Colistimethate	Microbiological assay of antibiotics (agar diffusion assay) Ph. Eur.
Budesonide	RP-HPLC: stability-indicating high performance liquid chromatography with ultraviolet detection
Fluticasone-17-propionate	RP-HPLC
Ipratropium bromide	RP-HPLC
Albuterol	RP-HPLC

2.3. Literature search

In order to get additional information about the compatibility/stability of mixed nebulizer solutions/suspensions for inhalation therapy we searched the literature databases "Medline" and "International Pharmaceutical Abstracts" for combinations of the terms "nebulizer solution", "inhalation", "aerosol", "compatibility", "stability", "combination", "HPLC", and "aerodynamic behavior".

3. Results and discussion

Both, the results of our own experimental studies and of the literature search are presented together for the respective nebulizable drug mixtures. (In)compatibility results for the eight most commonly used drugs in inhalation therapy are finally summarized in table format (Table 4). Further we introduce particular tripartite and quadripartite mixtures analyzed for physico-chemical compatibility and discuss the impact of certain excipients commonly used to formulate nebulizer solutions, i.e. preservatives and stabilizers. Finally, we present results from our aerodynamic studies performed with cascade impaction.

3.1. Dornase alfa

Dornase alfa (Pulmozyme®) is a mucolytic drug substance that decreases viscoelasticity and adherence of sputum, improves lung function and reduces respiratory exacerbations in cystic fibrosis (CF) patients. Administration is usually carried out via jet nebulizers, but a recent study indicates that perforated vibrating membrane devices deliver the drug more efficiently [3].

CF patients often mix dornase alfa with bronchodilatators like ipratropium bromide (Atrovent® and Atrovent® Fertiginhalat) and albuterol (Sultanol® and Sultanol® forte Fertiginhalat). Our studies

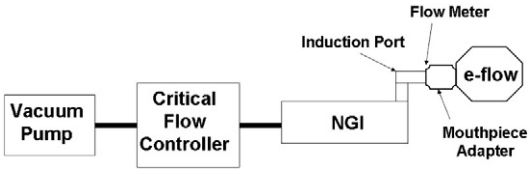

Fig. 1. Schematic test set-up of the impaction experiments.

Table 2
Results of the HPLC assays of quadripartite mixtures containing Colistin CF, Atrovent, Sultanol forte and Flutide forte [17].

n = 9	Ipratropium bromide	Salbutamol sulfate	Fluticasone-17-propionate
Nominal concentration [μg/ml]	55.55	26.66	22.22
Arithmetic mean ± relative SD [μg/ml]	58.4 ± 0.55	27.68 ± 0.35	23.82 ± 0.32
Percentage rate of nominal concentration	105.13	103.80	107.20

revealed that dornase alfa activity is affected by benzalkonium chloride, used as excipient in Atrovent® and Sultanol®, and disodium edetate used as an excipient in Atrovent®. But in the mixtures of Pulmozyme® with the preservative-free single dosage forms of Atrovent® 500 mg/2 mL Fertiginhalat or Sultanol® forte 2.5 mg/2.5 mL Fertiginhalat a decrease of dornase alfa activity was hardly measurable. Thus, these mixtures are most probably compatible [20]. To ensure this designation a more precise assay for determination of dornase alfa activity has to be applied and the nebulization properties of the proposed inhalation mixture have to be studied. Correspondent experimental studies were later on performed for mixtures of dornase alfa and tobramycin (see following paragraph). Ipratropium bromide and albuterol concentrations were not affected by mixing the drug products [20].

Concomitant inhalation of dornase alfa and antibiotics seems to be advantageous for antipseudomonal treatment [24] and thus, in daily practice many CF patients mix the drug with antibiotics, mainly tobramycin, which is available on the market in different formulations manufactured by different pharmaceutical companies. We analyzed activity and stability of dornase alfa after it was mixed with Tobi®, Bramitob® or Gernebcin®. We found no physical incompatibilities or decomposition in mixtures with Tobi® and Bramitob® [21], whereas the mixture with Gernebcin® is designated as incompatible. Results indicated that dornase alfa activity was especially affected by sodium metabisulfite used as an excipient in Gernebcin® (not in Tobi® and Bramitob®), since the activity loss was most pronounced when Pulmozyme® was mixed with pure solutions of sodium metabisulfite [18]. Tobramycin concentrations were not affected in any of the mixtures.

Mixed inhalation solutions of dornase alfa and the corticosteroid fluticasone-17-propionate (Flutide® forte Fertiginhalat) are considered as incompatible. Enzymatic activity of dornase alfa measured at room temperature declined from initially 93% of nominal activity to 72% after 24 h, probably due to the phosphate buffer used in Flutide® forte. Dornase alfa activity was hardly impaired in mixtures with the corticosteroid budesonide (Pulmicort®). Enzymatic activities measured up to a 31 hour interval after mixing resulted in at least 86% of the nominal activity and the mixtures were designated as compatible [25].

Table 3
Nebulizer and aerosol characteristics of Pulmozyme® mixed with 0.9% NaCl solution, Bramitob® or Tobi®. Results expressed as mean ± SD of triplicate assays of 4 test solutions [21].

	Pulmozyme® mixture with		
n = 12	0.9% NaCl solution	Bramitob®	TOBI®
Nebulization time [min] ± SD	5.85 ± 0.10	5.46 ± 0.22	5.66 ± 0.33
Delivered dose [%] ± SD	73.91 ± 2.78	74.41 ± 3.97	76.56 ± 2.62
MMAD [μm] ± SD	4.75 ± 0.13	4.73 ± 0.17	4.70 ± 0.08
GSD ± SD	1.65 ± 0.06	1.60 ± 0.00	1.63 ± 0.05
FPF < 5 μm [%] ± SD	54.36 ± 2.5	55.14 ± 2.5	54.70 ± 1.72

3.2. Antibiotics

3.2.1. Tobramycin

We investigated the compatibility of tobramycin and colistimethate nebulizable drug admixtures although we are aware that at present simultaneous inhalation is not an accepted standard in cystic fibrosis patients. However, mixing both antibiotics is practiced by patients, especially for treatment of mucoid *Pseudomonas aeruginosa* infections [26]. If antibiotic combination therapy via inhalation becomes clinically relevant, the physico-chemical compatibility of tobramycin and colistimethate is one of the prerequisites for simultaneous inhalation. Our study revealed that neither colistimethate nor tobramycin showed any losses of antibiotic potency after mixing. Values of pH and osmolality remained unchanged [27].

Physico-chemical compatibility of tobramycin (Nebcin®, 40 mg/mL) and albuterol (Ventolin®) has been proven earlier [28]. Our compatibility studies concerning mixtures of tobramycin (Tobi® and Gernebcin®) with the corticosteroids fluticasone-17-propionate (Flutide® forte Fertiginhalat) or budesonide (Pulmicort®) revealed that eight hours after mixing tobramycin concentrations remained unchanged, i.e. at a minimum of 96% of the nominal concentration [25].

3.2.2. Colistimethate

Pulmonary infections with Gram-negative bacteria resistant to tobramycin or other antibiotics in CF patients may require the inhalation of colistimethate in addition to standard inhalation therapy. Thus, we tested the physico-chemical compatibility of a quadripartite mixture containing colistimethate (Colistin CF) and the anticholinergic ipratropium bromide (preservative-free dosage form of Atrovent®), the β_2-agonist albuterol (preservative-free dosage form of Sultanol® forte), and the corticosteroid fluticasone-17-propionate (preservative-free Flutide® forte). Based on the results (see Section 3.9) we assume that incompatibilities in duplicate and triplicate mixtures containing colistimethate and one or two of the tested substances are unlikely. But results apply only to the preservative-free single dose formulations of these drugs [17].

Compatibility of colistimethate (Colistin® CF) and the glucocorticoid budesonide (Pulmicort®) was also tested. The duplicate mixture showed no loss in drug concentration of budesonide and no change in antibiotic activity of colistimethate over a period of 24 h after mixing. During the first six hours after mixing an increase of the pH (5.5 to 7.3) was measured [22] and according to the definition of compatibility the mixture is to be categorized as incompatible. But because no other parameters, i.e. osmolality and physical appearance, were altered and chemical stability of the active substances was given over a period of 24 h,

Table 4
Physico-chemical compatibility of inhalation solutions/suspensions. No sufficient information available for yellow marked combinations. *Mixtures not recommendable from a clinical viewpoint. **Compatibility applies only to preservative-free dosage forms. #Unchanged aerosol characteristics and drug output have been proved.

	Dornasealfa Pulmozyme®	Tobramycin Bramitob® TOBI®	Tobramycin Gernebcin®	Colistimethate Colistin CF®	Ipratropium Atrovent®, Atrovent®unit dose 2 ml	Albuterol Sultanol®, Sultanol®unit dose 2.5 ml	Budesonide Pulmicort®	Fluticasone–17–propionate Flutide®	Cromolyn Intal®	Hypertonic saline 5.85% NaCl solution
Dornasealfa		Mixable#	Do not mix	Do not mix	Do not mix	Do not mix	Mixable	Do not mix	Do not mix	Do not mix
Tobramycin Bramitob®,TOBI®	Mixable#			Mixable	Mixable	Mixable	Mixable	Mixable	Do not mix	Do not mix
Tobramycin Gernebcin®	Do not mix			Mixable	Mixable	Mixable	Mixable	Mixable	Do not mix	Do not mix
Colistimethate	Do not mix	Mixable	Mixable		Mixable**	Mixable**	Mixable	Mixable	Do not mix	Mixable
Ipratropium	Do not mix	Mixable	Mixable	Mixable**		Mixable#	Mixable	Mixable#	Mixable**	Do not mix
Albuterol	Do not mix	Mixable	Mixable	Mixable**	Mixable#		Mixable	Mixable#	Mixable**	Do not mix
Budesonide	Mixable	Mixable	Mixable	Mixable	Mixable	Mixable		*	Mixable	Mixable
Fluticasone–17–propionate	Do not mix	Mixable	Mixable	Mixable	Mixable#	Mixable#	*		Do not mix	Do not mix
Cromolyn	Do not mix	Do not mix	Do not mix	Do not mix	Mixable**	Mixable**	Mixable	Do not mix		Do not mix
Hypertonic saline	Do not mix	Do not mix	Do not mix	Mixable	Do not mix	Do not mix	Mixable	Do not mix	Do not mix	

we judged mixtures of Colistin® CF and Pulmicort® to be physico-chemically compatible [22].

We also studied compatibility of admixtures containing colistimethate and 5.85% sodium chloride solution, since nebulized hypertonic saline solution has been established as an effective adjunctive therapy for respiratory symptoms in CF patients [29,30]. We found the mixture to be physico-chemically compatible and stable over a period of 48 h stored under refrigeration [17].

3.3. Ipratropium bromide

We evaluated the physico-chemical compatibility of the anticholinergic ipratropium bromide (Atrovent® LS), the β_2-agonist albuterol (Sultanol®) and the corticosteroid fluticasone-17-propionate (Flutide® forte) by experimental studies of the tripartite mixture with validated analytical assays (see Section 3.9). We found compatibility over a period of five hours, i.e. almost no changes in drug concentrations, pH and osmolality could be detected and no visible changes occurred [19]. Thus, we consider the duplicate mixtures of these drug products to be compatible.

Compatibility of different formulations of ipratropium bromide (Atrovent®, Duovent®, i.e. fenoterol plus ipratropium) and budesonide (Pulmicort®) were tested by different working groups. No evidence of any physico-chemical incompatibility was found [10,31,32].

When ipratropium bromide and cromolyn (Intal®) containing inhalation solutions were mixed compatibility was given only with the preservative-free formulation of Atrovent® [10,33–35].

Finally, a study from China raised concerns about the packaging material of ipratropium bromide containing aerosol products. Yue et al. [36] reported that the active substance is absorbed and leachable additives from the primary package may be present in the drug solution.

3.4. Albuterol and levalbuterol

In Europe the most common β_2-agonist prescribed is albuterol, but its stereoisomer levalbuterol is widely used in the US. Both substances have similar pharmacokinetic and pharmacodynamic properties. We concentrated on albuterol whereas in one recently published study the compatibility of different duplicate mixtures containing levalbuterol was evaluated.

3.4.1. Albuterol

In several studies the compatibility and stability of mixtures of albuterol and ipratropium bromide was proven [19,35,37]. Moreover, there are combination products for nebulization available which contain both albuterol and ipratropium bromide, e.g. Berodual® LS in Germany and DuoNeb® in the US. For these products stability is proven.

Several studies proved the compatibility of albuterol (Proventil® or Xopenex®) and budesonide (Pulmicort®) [10]. Recently Melani [38] tested the aerosol characteristics of this particular drug mixture (see Section 3.11.).

The mixture of albuterol (Ventolin®) and cromolyn (Intal®) was also found to be compatible [10,39].

3.4.2. Levalbuterol

Bonasia et al. [40] investigated several two-drug admixtures of levalbuterol by visual inspection, pH measurement and high performance liquid chromatography (HPLC assays). For duplicate nebulizable drug admixtures containing levalbuterol and budenoside, ipratropium bromide, cromolyn (DNCG), or acetylcysteine sodium no evidence of physical incompatibility and/or chemical decomposition were registered over a period of at least 30 minute storage at room temperature.

Yamreudeewong et al. evaluated the stability of a mixture containing levalbuterol and ipratropium over a period of 28 days at room temperature. They found no significant decrease in concentrations of the active drugs, i.e. decrease in concentration of each drug was below 10% of the initial concentration [41].

3.5. Budesonide

Compatibility of budesonide and cromolyn is known [10,31,42]. When budesonide nebulizable suspension (Pulmicort®) was mixed with 5.85% sodium chloride solution no loss in drug concentration of budesonide was detected over a test period of 24 h. Osmolality and pH remained unchanged and visual inspection revealed no abnormality [22].

3.6. Cromolyn

Although cromolyn is not a standard therapy in cystic fibrosis we decided to report compatibility with other nebulizable drug products. As stated before [10], 7% of patients at our CF center reported inhaling cromolyn mixed with other drugs and prescribing information about miscibility with cromolyn is not consistent. Information about compatibility is reported in the above Sections 3.3, 3.4.1, and 3.5.

3.7. Hypertonic sodium chloride solution

We determined the physico-chemical compatibility of 5.85% sodium chloride (NaCl) solution with either colistimethate (see Section 3.2.2) or budesonide (see Section 3.5). Fox et al. [43] assessed the physical compatibility of eleven nebulizable drug products with 7% sodium chloride solution. The duplicate mixtures of 7% NaCl solution and acetylcysteine, albuterol, atropine, cromolyn sodium, dexamethasone, glycopyrrolate, ipratropium, metaproterenol, sodium bicarbonate, terbutaline, or tobramycin were analyzed by serial turbidimetric testing and visual inspection. The authors stated compatibility for all different mixtures, except for cromolyn sodium, which was found to be visually incompatible with 7% NaCl solution.

3.8. Miscellaneous

Formoterol and arformoterol were analyzed by Akapo et al. [11] with regard to physico-chemical compatibility and aerodynamic characteristics (see Section 3.11) of different duplicate mixtures. They found compatibility for formoterolfumarate 20 mg/2 ml when mixed or sequentially nebulized with budesonide inhalation suspension 0.5 mg/2 ml, ipratropium bromide 0.5 mg/2.5 ml, cromolyn sodium 20 mg/2 ml, or 10% acetylcysteine solution (100 mg/ml). However, mixing or sequential nebulization significantly increased the amount of drug delivered compared with single drug nebulization and the authors emphasize that clinical implications of the in vitro data have not been determined.

The chemical and physical compatibility of duplicate mixtures of arformoterol (15 mg/2 ml) with ipratropium bromide (0.5 mg/2.5 ml), acetylcysteine (800 mg/4 ml), or budesonide (0.25 mg/2 ml and 0.5 mg/2 ml) was tested. Visual inspection, pH measurement, and HPLC assays of each active component revealed no signs of incompatibility in any of these mixtures [44].

3.9. Tripartite and quadripartite mixtures

We analyzed several tripartite mixtures containing the anticholinergic ipratropium bromide (Atrovent® LS) and the β_2-agonist albuterol (Sultanol®). Physico-chemical compatibility was proven with fluticasone-17-propionate (Flutide® forte) [19]. Mixtures of albuterol, tobramycin and ipratropium bromide (no information available about studied brands) were found to be compatible, too [10].

In mixtures with cromolyn (Intal®) compatibility is given for mixtures prepared with the preservative-free single use dosage forms Atrovent® Fertiginhalat and Sultanol® forte Fertiginhalat [34]. Compatibility seemed to be affected by certain excipients like benzalkonium chloride. In order to minimize the risk of physical incompatibility, only preservative-free formulations of albuterol or ipratropium bromide inhalation solutions should be mixed with Intal® [10,34].

Comparable results were obtained after mixing ipratropium bromide and albuterol with dornase alfa (Pulmozyme®). Dornase alfa activity was more compromised in mixtures containing the excipients benzalkonium chloride and disodium edetate. Thus, the preservative-free single use dosage forms Atrovent® Fertiginhalat and Sultanol® forte Fertiginhalat are designated as mixable with Pulmozyme® without reducing the efficacy of dornase alfa [20].

The compatibility of Berodual® and Pulmicort® was tested in an experimental setting. The tripartite mixture, i.e. Berodual® which contains ipratropium bromide and fenoterol, turned out to be physico-chemically compatible [10,34].

We analyzed the physico-chemical compatibility of colistimethate (Colistin CF), fluticasone-17-propionate (Flutide® forte), ipratropium bromide and albuterol. This quadripartite mixture was found to be compatible, but only when preservative-free single-dose formulations of the drug products (Atrovent® Fertiginhalat, Sultanol® forte Fertiginhalat) were used (Table 2).

3.10. Excipients

Many multiple dosage forms of inhalative drugs contain not only the active agent but also excipients as preservatives and stabilizers, e.g. benzalkonium chloride, disodium edetate, and sodium metabisulfite. Our studies revealed that most probably these additives are responsible for detected incompatibilities, e.g. limited activity of dornase alfa (Pulmozyme®) and colistimethate

(Colistin® CF). Hence, patients should be informed about possible incompatibilities caused by excipients and should be educated to check the excipients declared on the package and package insert of each medicinal product.

3.11. Aerosol characteristics

Aerosol output of the nebulizer and particle size distribution of the aerosol produced are the most important issues for successful inhalation therapy, since particle size influences the deposition pattern and bioavailability of drugs delivered to the respiratory system [45]. Special conditions are required for an effective inhalation therapy of infants and young children [46]. To figure out if mixing of dornase alfa (Pulmozyme®) and tobramycin (Tobi® and Bramitob®) causes changes in particle size distribution, the physico-chemical compatible admixtures (see Section 3.1) were nebulized with the PARI eFlow® rapid. Aerosol characteristics, i.e. MMAD, GSD and FPF, of dornase alfa were assessed via cascade impaction with the NGI. SE-HPLC was used to assess the dornase alfa concentrations deposited on the impactor stages. The aerosol patterns of admixtures of dornase alfa and tobramycin were compared to those of dornase alfa diluted with 0.9% sodium chloride solution in order to achieve the same volume. There was no difference in particle size distribution, MMAD, GSD and FPF (Table 3).

Melani [38] evaluated the aerosol output, drug output, and aerosol particle size characteristics of two corticosteroids (budesonide, beclomethasone dipropionate) and two bronchodilators (albuterol, ipratropium bromide) when nebulized alone or in tripartite mixtures (budesonide or beclomethasone and albuterol and ipratropium).

Using the SideStream and VentStream-Pro nebulizers, run with the AirClinic compressor, the author found that mixing reduced drug output and increased mass median aerodynamic diameter (MMAD) with the SideStream, but not always with the VentStream-Pro. However, drug output and MMAD never reached critical values, i.e. outside the respirable range. When nebulized alone, the respirable mass of bronchodilators ranged from 18% to 40% of the nominal dose compared to 13% to 37% when mixed. The respirable mass of corticosteroids nebulized alone ranged from 10% to 24% of the nominal dose, in mixtures it ranged from 10% to 17%. Hence, both nebulizers can be recommended in terms of aerosol performance for inhalation therapy with these particular drug mixtures.

Akapo et al. [11] assessed the aerodynamic characteristics of formoterol fumarate (20 mg/2 ml) when mixed or sequentially nebulized with budesonide inhalation suspension (0.5 mg/2 ml), ipratropium bromide (0.5 mg/2.5 ml), cromolyn sodium (20 mg/2 ml), or acetylcysteine 10% (100 mg/ml). Aerosols were generated with a Pari LC Plus® nebulizer and characterized by using an 8-stage cascade impactor. They found a significantly increased respirable fraction/delivered dose ($p < 0.05$) for the mixed or sequentially nebulized drugs and assume that this was due to an increased drug volume or reconcentration in the nebulizer cup by sequential nebulization. Both facts decreased the amount of drugs retained in the dead volume. In contrast, the FPF, MMAD, and GSD generally remained unchanged for all admixtures, except for the FPF of the formoterol/budesonide combination. This may be caused by polysorbate 80 used as surfactant in the budesonide formulation. The authors conclude that mixing or sequential nebulization significantly increases the amount of drug delivered compared with single drug nebulization.

Itazawa et al. [47] analyzed the aerosol characteristics of a duplicate mixture containing budesonide (Pulmicort®) and the β_2-agonist procaterol (Meptin®). They found no relevant differences in drug output and particle size distribution with regard to budesonide when nebulized alone or in the admixture. In contrast, they detected a significantly higher amount of delivered procaterol and a higher rate of smaller particle sizes containing procaterol in the admixture compared to the unmixed solution.

The physico-chemical compatible tripartite mixture containing fluticasone-17-propionate (Flutide® forte), albuterol (Sultanol® forte Fertiginhalat) and ipratropium bromide (Atrovent® Fertiginhalat) [19] was nebulized by Stanko and the aerosol characteristics were analyzed. Particle size distribution, MMAD, GSD, and FPF were assessed via cascade impaction and results revealed no relevant changes [34].

4. Conclusion

Our in vitro studies and search of relevant literature indicate that most of the tested nebulizer suspensions are mixable without provoking any measurable incompatibilities. A major exception is dornase alfa. Its activity was affected by certain excipients contained in some of the drug products. The same excipients caused incompatibility when albuterol and/or ipratropium bromide was mixed with cromolyn or colistimethate.

Mixtures should always be prepared directly before inhalation and surplus quantities should be discarded.

Only a limited number of mixtures have been tested yet with regard to aerosol characteristics. More studies assessing particle size distribution of common mixtures administered with popular nebulizers are needed.

Finally, the clinical efficacy of inhalation therapy with duplicate, tripartite or even quadripartite mixtures must be evaluated in comparison to consecutive inhalation of the same drugs, before final recommendations for patients can be made.

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